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A novel approach to the use of genetically modified herbicide tolerant crops for environmental benefit

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The proposed introduction of genetically modified herbicide tolerant (GMHT) crops, with claims of improved weed control, has prompted fears about possible environmental impacts of their widespread adoption, particularly on arable weeds, insects and associated farmland birds. In response to this, we have developed a novel weed-management system for GMHT sugar beet, based on band spraying, which exploits the flexibility offered by the broad-spectrum partner herbicides. Here, we show the results from two series of field experiments which, taken together, demonstrate that, by using this system, crops can be managed for enhanced weed and insect biomass without compromising yield, thus potentially offering food and shelter to farmland birds and other wildlife. These results could be applicable widely to other row crops, and indicate that creative use of GMHT technology could be a powerful tool for developing more sustainable farming systems in the future.

Keywords: genetically modified crops; biodiversity; sustainability; sugar beet; weeds

1. INTRODUCTION

Sugar beet is a poor competitor with weeds in arable fields because it is slow growing early in the season and has a low canopy in its first year of a biennial life cycle. Good weed control is therefore essential to produce economically viable yields (Jansen 1972), but is not easy to achieve with current selective herbicides and/or inter-row tillage. From the point of view of competition for resources (principally light), weed control need not be carried out until the six to eight leaf growth stage of the crop (Scott *et al.* 1979), but the weaknesses of current conventional herbicides dictate that weed control commences pre-emergence or at the cotyledon stage of weeds (and crop). Thus very few weeds are present throughout the season in most crops. However, the few crops that are weedy do offer a food source for migrating seed-eating birds in the autumn (Wilson *et al.* 1999; Watkinson *et al.* 2000). It is the potential loss of these weedy crops, amid general alarm over population decline of farmland bird species (Chamberlain *et al.* 2000), which has prompted concerns about genetically modified herbicide tolerant (GMHT) technology in the UK from English Nature (English Nature 1998, 2000) and some environmental scientists (Krebs *et al.* 1999; Hails 2000) and non-governmental organizations.

However, our novel approach to weed management in GMHT sugar beet exploits the much greater flexibility and efficacy of the broad-spectrum herbicides, glyphosate and glufosinate–ammonium, to which GM tolerances have been produced. We have developed a simple over-the-row band-spraying technique to control in-row weeds first, whilst those between the rows can be controlled by a later overall spray application. This exploits both the temporal and spatial flexibility offered by the GMHT system, to allow weed control that is tailored precisely to the avoidance of competition.

Two series of experiments have tested the hypothesis that, in GMHT systems, weed-management options could be found that would benefit weed and invertebrate populations of relevance to farmland birds without reducing crop yield.

2. METHODS

A first series of five experiments investigated the effect of weed-management strategies on the yield of glyphosate-tolerant GM sugar beet (L 77 from Monsanto) in East Anglia, UK in 1999 and 2000. The experiments were carried out on soil types typical of those on which sugar beet is grown in the UK. Between 12 and 22 weed species were present in each experiment, many of them important components of farmland bird diets (Krebs *et al.* 1999). *Chenopodium album* was an important weed on four sites, *Fallopia convolvulus* and *Veronica persica* on three, *Sinapis arvensis*, *Persicaria maculosa*, volunteer cereals (from previous crop), *Cirsium arvense* on two and *Tripleurospermum maritimum*, *Persicaria lapathifolia* and *Alopecurus myosuroides* on one. Total population densities in untreated plots ranged from 29 to 75 m^{-2}.

The conventional herbicide programmes varied at each site depending on the weed species present. The number of active ingredients ranged from three to eight, the simplest including phenmedipham, metamitron and ethofumesate (site 1, 1999), and the most complex including the above three plus paraquat, diquat, desmedipham, lenacil and cycloxydim. Other active

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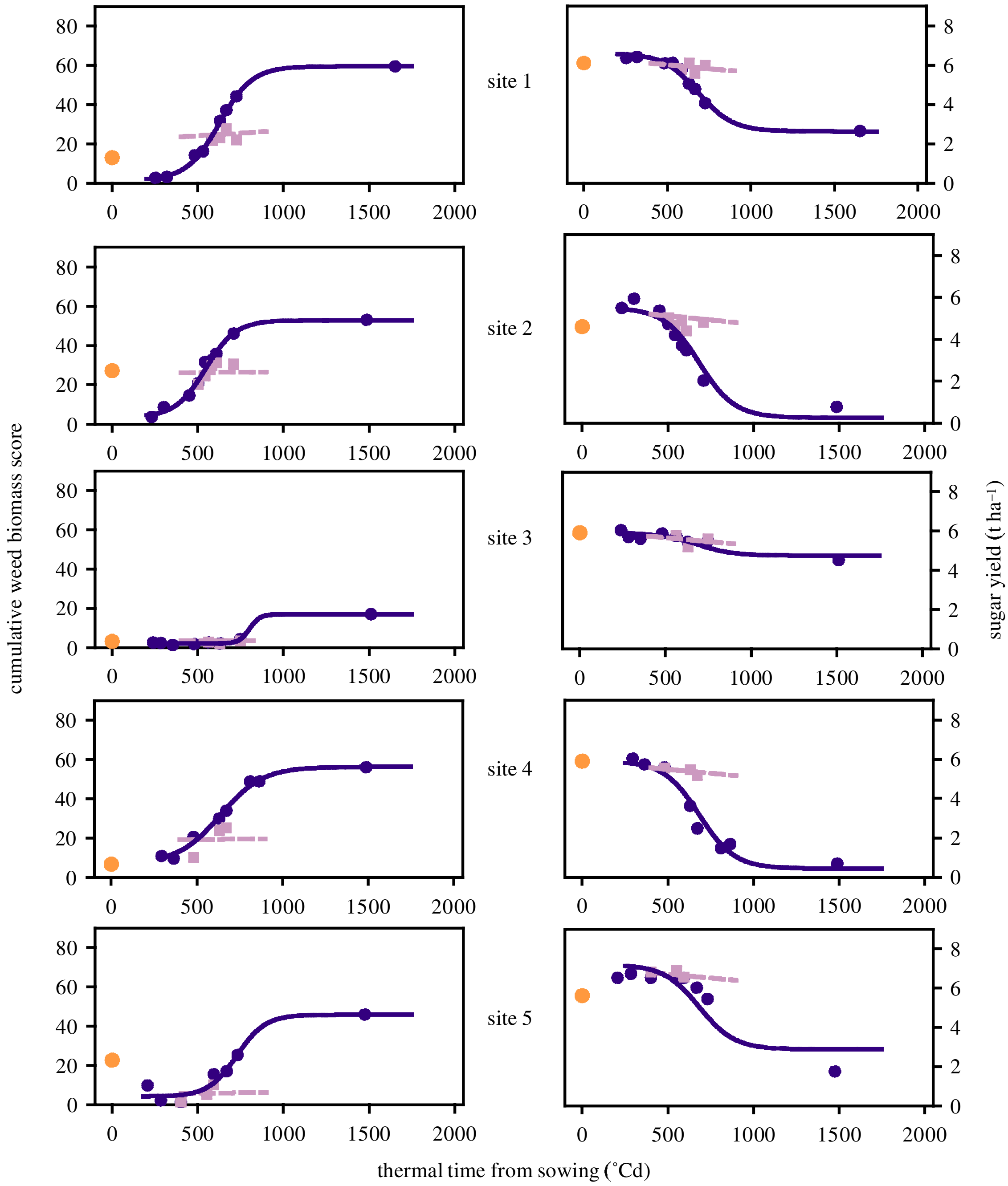

Figure 1. Effects of the timing (°Cd after sowing) of the first overall sprays of glyphosate in a two spray regime (blue circles) and the second overall spray following a band spray (pink squares) on the cumulative biomass of weeds (left column figure) and sugar yield of sugar beet (right column figure) at five sites in 1999 and 2000. Data for conventional herbicides are given by orange circles.

ingredients used on the other sites included clopyralid and triflusulfuron-methyl. The number of applications ranged from two to four. In 1999, applications of some treatments, particularly in the conventional programmes, were delayed by adverse weather conditions. Treatments of glyphosate (at 1080 g active ingredient (a.i.) sprayed ha^{-1}) were applied either overall at several timings between 207 and 864 day degrees (°Cd) above 3 °C after sowing, or over the sugar beet rows only at similar but fewer timings up to 586 °Cd. The overall treatments were followed by a second application between 698 and 1022 °Cd and the band sprays by an overall application between 401 and 811 °Cd, both depending on the timing of the first sprays. Glyphosate treatments were compared with untreated controls and programmes of current commercial herbicides applied pre-emergence (in two trials), but mostly post-emergence starting between 79 and 222 °Cd.

The biomass of weeds present in each treatment was assessed on six occasions throughout the season, the earliest at the time of the first glyphosate applications in late May (240 °Cd) and the latest in mid-August (1450 °Cd). Biomass was assessed using scores on a linear scale (0–10), where 0 represents no living weeds and 10 represents full biomass for the time of year and plant stage with no effect on plants. In band-sprayed plots, the score was a mean of the sprayed area down the row and unsprayed area between the row. Where weed numbers were low, scores in untreated plots were sometimes less than 10. Sugar yield was assessed at harvest in late August/early September. This was earlier than commercial crops due to the con-

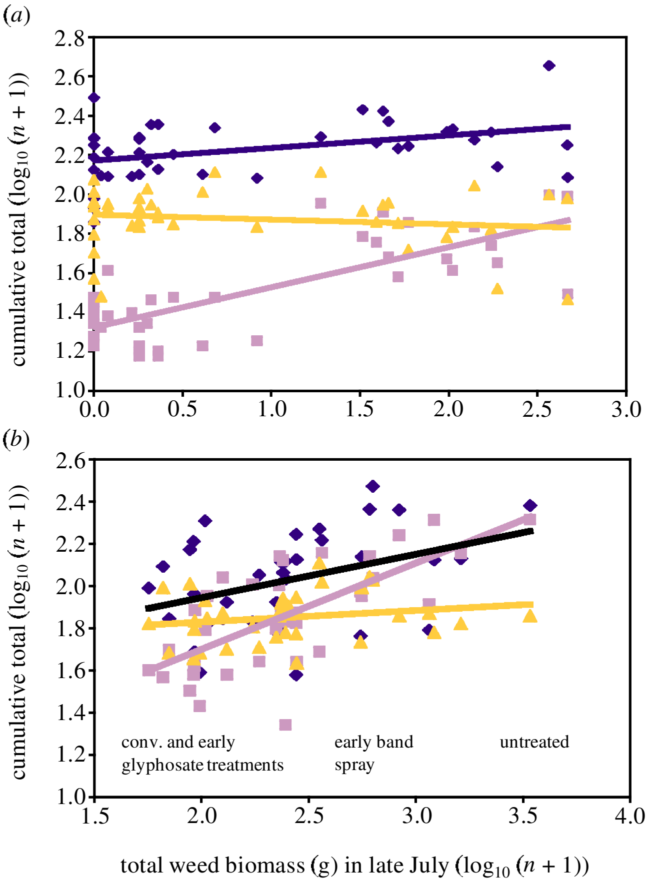

Figure 2. Effect of weeds on abundance of arthopods in GMHT sugar beet. (Blue diamonds, carabids; pink squares, staphylinids; orange triangles, spiders.) (*a*) Carabids $= 0.0641x + 2.1717$, $r^2 = 0.15$; staphylinids $= 0.204x + 1.323$, $r^2 = 0.59$; spiders $= -0.0246x + 1.8961$, $r^2 = 0.02$. (*b*) Carabids $= 0.2055x + 1.5352$, $r^2 = 0.14$; staphylinids $= 0.4089x + 0.8831$, $r^2 = 0.47$; spiders $= 0.0547x + 1.7212$, $r^2 = 0.04$.

straints of the consent and the audit requirements of British Sugar plc.

The environmental impact of all the conventional herbicide programmes was assessed by the Millieumeetlat scoring system (CLM 2000), which evaluates toxicity, mobility and persistence of pesticides based on data from submissions for the registration of pesticides in Europe.

In a second series of four experiments using the same cultivar and husbandry, a subset of the treatments included in the yield experiments was set up in adjacent areas at three (sites 1, 3 and 4) of those five sites, and one at a different site (site 6). Plot sizes were larger to allow the collection of invertebrates with reduced interference from neighbouring plots. For this study, we selected Carabidae, Staphylinidae and Aranae, sampled in pitfall traps, as indicators of arthropod abundance. Pitfall traps give a measure of abundance and activity and are widely used to study ground-dwelling invertebrates (Baars 1979). Carabidae are regarded as useful environmental indicators (Luff & Woiwod 1995), and both staphylinids (Powell *et al.* 1985) and spiders (Haughton *et al.* 1999) contain species that are known to respond to herbicide regimes. Traps were set for 7 days per sampling occasion in 1999, and for 14 days per occasion in 2000 to increase sample size. Samples have to be standardized for number of individuals for meaningful comparisons of diversity to be made. Here, we used the log-series α diversity parameter for reasons given by Taylor *et al.* (1976).

3. RESULTS

(a) *Weed management and yield effects*

Cumulative weed biomass in untreated plots at four of the five sites was high, but low at site 3 in 2000, where weed numbers were lowest. All treatments at all sites significantly reduced weed numbers and biomass compared with the untreated plots, but weed control from the early overall glyphosate programmes was generally better than that from the conventional treatments, particularly at sites 2 and 5 (figure 1). Weed biomass was greater than conventional following the later overall sprays of glyphosate. Biomass in band-sprayed plots was much higher between rows than was evident from the scores.

Sugar yields from all trials were lower than would normally be expected from a commercial crop (range 4.9–6.1 tonnes per hectare ($\mathrm{t\,ha^{-1}}$) in plots treated with conventional herbicides) as a result of the imposed early harvest in late August or early September to comply with British Sugar GM audit requirements. Yield reductions in the untreated plots compared with those treated with conventional herbicides ranged from 24 to 88%. Glyphosate, first applied overall sometime between 240 and 320 °Cd, gave the best yields in each trial (range 5.9–6.7 $\mathrm{t\,ha^{-1}}$)—on average 9.7% greater than the conventional treatments, although the differences were only significant at sites 2 and 5 (figure 1). In previous trials, yield reductions ranging from 5–15% in conventional treatments compared with the best glyphosate treatment (Moll 1997; Brants & Harms 1998; Wilson 1999; Wilson *et al.* 2002) have been attributed partially to the slight, but occasionally important, phytotoxic effects of conventional herbicides on the sugar beet plants themselves (Wilson *et al.* 2002). In our experiments, some of the yield improvement from the glyphosate treatments was probably also a result of better weed control throughout the season.

Delays in glyphosate treatment had a significant effect on final sugar yield (table 1; figure 1), described by the following equation:

$$Y = Y_0 + \alpha/(1 + e^{\beta(X - X_0)}), \tag{3.1}$$

where Y is the sugar yield, Y_0 is the yield from the untreated plots, α is the maximum reduction of sugar yield observed and β is the rate of yield reduction due to delays in treatment. $Y_0 + \alpha$ combine to represent the maximum obtainable sugar yield when weeds are effectively controlled to the full, X_0 is the thermal time (°Cd) at which the reduction of sugar yield is at half value of α, and X is the thermal time from sowing. When α and Y_0 were allowed to vary from experiment to experiment, but the other parameters were fixed, the total variance accounted for (r^2) was 97.1% (d.f. = 34).

The yields from the band-spray treatments could be described by a simple linear relationship:

$$Y = Y_p - \gamma X, \tag{3.2}$$

where Y is the sugar yield, Y_p is the intercept indicating the potential yield at a given site in a given year, γ is the slope measuring the reduction of sugar yield per unit of delay in thermal time from sowing, and X is the thermal

Table 1. The estimates and their standard errors (s.e.) of parameters in (*a*) equation (3.1) describing yields from the overall sprayed treatments and (*b*) equation (3.2) describing yields from the band-sprayed treatments.

(*a*) site	Y_0 (s.e.)	X_0 (s.e.)	α (s.e.)	β (s.e.)
1	2.6274 (0.4085)	683.4 (19.27)	3.9745 (0.5624)	0.0097 (0.0019)
2	0.2702 (0.4321)	683.4 (19.27)	5.2385 (0.5730)	0.0097 (0.0019)
3	4.7461 (0.3941)	683.4 (19.27)	1.1459 (0.5133)	0.0097 (0.0019)
4	0.4385 (0.3802)	683.4 (19.27)	5.4711 (0.5867)	0.0097 (0.0019)
5	2.8701 (0.4104)	683.4 (19.27)	4.3118 (0.5492)	0.0097 (0.0019)

(*b*) site	Y_p (s.e.)	γ (s.e.)
1	6.4135 (0.4291)	−0.0008 (0.0005)
2	5.9190 (0.4836)	−0.0008 (0.0005)
3	6.0546 (0.4315)	−0.0008 (0.0005)
4	5.8842 (0.3998)	−0.0008 (0.0005)
5	7.1755 (0.3531)	−0.0008 (0.0005)

Table 2. The effects of herbicide treatments on abundance and biodiversity of carabids, staphylinids and spiders caught (cumulative total) in pitfall traps in selected treatments at each of four sites.
(*n*, number of carabids + staphylinids + spiders per treatment (all 12 traps); s.e., standard error; e, early; m, mid-timing; l, late; ll, later; b, band.)

			treatment						
site	sample weeks	parameter	untreated	conventional	glyphosate e + l	glyphosate m + l	glyphosate l + ll	glyphosate eb + l	total (all treatments)
1 (1999)	4	*n*	3346	2459	2353	2536	3131	—	12 732
		no. of species	48	46	47	48	50	—	
		α index	7.94	8.03	8.32	8.40	8.45	—	9.80
		s.e. of α	1.25	1.30	1.34	1.33	1.31	—	1.26
6 (1999)	4	*n*	2752	2493	2402	2525	2646	—	12 852
		no. of species	37	37	33	35	40	—	56
		α index	6.04	6.16	5.41	5.75	6.69	—	7.52
		s.e. of α	1.09	1.11	1.03	1.06	1.16	—	1.08
3 (2000)	9	*n*	3796	3403	3113	3690	3207	3528	31 911
		no. of species	63	50	50	52	53	58	86
		α index	10.73	8.31	8.46	8.57	9.02	9.86	10.80
		s.e. of α	1.48	1.29	1.31	1.30	1.36	1.42	1.24
4 (2000)	7	*n*	1694	894	755	796	943	781	9889
		no. of species	48	41	42	34	38	36	68
		α index	9.19	8.87	9.59	7.22	7.94	7.80	9.83
		s.e. of α	1.47	1.56	1.68	1.39	1.45	1.47	1.29

time delay from sowing. Comparison of regressions from all sites showed that each had a different Y_p but a common slope γ, which was not significantly different from zero, and accounted for 90.2% of the total variance (d.f. = 18) in the observed sugar yields (table 1; figure 1). Delays in overall sprays following band treatments resulted in the same amount of sugar yield reduction per unit of thermal time in each trial.

These results indicate that weed control with overall glyphosate applications should commence at around 275 °Cd for optimum yield return and before 535 °Cd if significant yield loss is to be avoided. This broadly agrees with previous work (Scott *et al.* 1979; Schweizer & Dexter 1987; Wilson *et al.* 2002). Results from the band-spray treatments indicate that, following a first spray applied between 207 and 530 °Cd after sowing, the second could be applied much later between 586 °Cd and 725 °Cd (average 656 °Cd) before significant reductions in yield compared with the conventional regime occur.

(b) *Environmental impact*

In our assessment, using the Millieumeetlat system of the direct environmental impact of the herbicide regimes, scores for the conventional herbicides ranged from 32 to 218 for water organisms, 11 to 960 for soil organisms and 155 to 16 540 for deeper water. The equivalent scores for glyphosate treatments were 0, 5–6 and 0, respectively, even though the latter used the maximum dose recommended on draft labels, and conventional treatments, especially in 1999, were less intensive compared with most commercial treatments used in that season as a result of the later sowing. A score greater than 100 is considered unacceptable for an individual application in the Millieumeetlat system. All herbicides were within the acceptable limits for water organisms, but lenacil in experiments 2, 3 and 5 and clopyralid in experiment 4 were above this limit for deeper water, and lenacil (experiments 2 and 5) and paraquat plus diquat (experiments 4 and 5) were above this limit for soil organisms.

(c) *Effects on arthropods*

In our study sites, the number of species of carabids, staphylinids and spiders was typical of arable fields (Kromp 1999). Carabids were more numerous than the other two groups at all sites, comprising at least 44% of the total collected (site 4), but as high as 84% at site 6, which was situated next to a beetle bank. Staphylinids comprised between 5% (sites 6 and 3) and 30% (site 4), while spiders comprised between 10% (site 6) and 31% (site 1) of the total. Site 3 was the most diverse (particularly in spiders), and caught the largest number of specimens over the sampling period (31 911) even taking account of the longer period of collection (table 2). Site 1 had moderate populations, while site 4 had the fewest individuals.

Among the carabids, the dominant species at all sites was *Pterostichus melanarius*, which comprised at least 70% of the carabids. The Aleocharinae were the dominant group of staphylinids at two of the four sites (6 and 3), constituting 79 and 50% of the populations there respectively. *Philonthus cognatus* was the most important staphylinid species at the other two sites, making up 43% at site 1 and 53% at site 4. Spider communities were also dominated by a single species at three of the four sites. *Oedothorax apicatus* was the most common species at sites 1 (61%), 6 (49%) and 3 (69%), but *Erigone atra* was dominant at site 4 (36%). In all three groups, the top five species made up at least 87% of the total at any site.

The impact of herbicide treatments on the relative abundance of the three groups depended on the density and diversity of weeds present and the timing and efficiency of their removal. At sites 3 and 6, there was no consistent effect of treatments on the numbers of carabids, staphylinids or spiders at any time during the growth of the crop, only an occasional transitory effect. This was almost certainly due to the low weed populations at these sites (*ca.* 11–12 m^{-2} in untreated plots), which did not alter the structure of the habitat sufficiently to influence the populations of these arthropods. Indeed, the maximum ground cover afforded by those weeds in late July was only 23 and 16% respectively, in the untreated plots at the two sites, compared with 35% at site 1 and 96% at site 4.

At these latter two sites, weed numbers in untreated plots were two to five times greater (27 and 61 m^{-2}, respectively) and the weeds, especially at site 4, were much taller. There were strongly significant correlations at both sites between weed biomass (including dead and dying weeds) in late July and the cumulative numbers of staphylinid beetles collected in the pitfall traps during the sampling period of June–August (figure 2). The correlation was much weaker, although still significant, for carabids but non-significant for spiders.

Thus fewest carabids and staphylinids were found in plots treated with conventional herbicides or early overall applications of glyphosate, and most where weed control was delayed (sites 1 and 4) or partial, as in the band-sprayed plots (site 4). There was no difference for any species of carabid or staphylinid, or their combined totals, on any sampling occasion or when considering cumulative totals, between the conventional treatments and the early overall glyphosate treatment. This indicates that the response of the beetles was to the removal of weeds, and not to the chemicals used.

These effects of herbicides were similar to those reported in cereal crops (Powell *et al.* 1985; Haughton *et al.* 1999; Moreby & Southway 1999). In one study carried out in a row crop, Purvis & Curry (1984) reported that carabids were rarely affected by weediness in sugar beet fields but staphylinid beetles were substantially increased, especially *P. cognatus*, while spiders were unaffected. Spiders are known to be affected by herbicide regimes but the response is more apparent in some families than others (Haughton *et al.* 1999).

As expected, there were site differences in invertebrate diversity. The most diverse was site 3 and the least, site 6, which was significantly less diverse. However, there were no significant differences between any of the other sites (table 2). Within any site there was no significant difference in the log series α index of biodiversity between any treatment on any one sampling date, or when the cumulative catch over all sampling dates was considered, even at the two sites that showed significant effects of treatments on the number of carabid and staphylinid beetles. There was no difference between the conventional or early-applied glyphosate herbicides. The lack of effects of treatments on biodiversity for individual sample dates is not surprising as any actual change in species complement would only be likely to occur over long periods for the relatively widespread and abundant farmland species being sampled, many of which have only one generation per year.

4. DISCUSSION

These experiments demonstrate that GMHT technology allows a flexible knowledge-based management approach to weed control in sugar beet, permitting higher weed populations early in the season than is possible in conventional systems. The models of yield effects described here could be used to determine weed control requirements quantitatively. In some low weed pressure situations, such as at site 3 (figure 1), only one well-timed spray would be needed to achieve satisfactory commercial weed control.

Inputs could be tailored to weed pressure and environmental objectives, such as weed-free fields for bird species such as the stonecurlew (*Burhinus oedicnemus*) or to provide low-growing vegetation for skylarks (*Alauda arvensis*), both species of current conservation concern in the UK. These and other scenarios, for example availability of weed seed late in the season, can be created by band spraying appropriately in GMHT sugar beet. Weeds can also help to minimize insecticide use by reducing colonization of beet by migratory insect pests, such as aphids, either offering alternative hosts, or providing olfactory and/or visual distraction (Dewar *et al.* 2000; Finch & Collier 2000). In addition, the avoidance of tractor hoeing in our GMHT weed-management systems also means that there is potential for improving the habitat for some species of ground-nesting farmland birds.

Concerns have long been expressed about the effects of intensification of agriculture on the farmland environment, most recently by Donald *et al.* (2001). Some of the immediate environmental issues about GMHT are being addressed based on current agronomic practice in the Government's major Farm Scale Evaluations (FSE) project, which examines one management option for GM crops (Firbank *et al.* 1999). Our work reported here examines a much wider range of weed-management options, albeit on a small scale and on a single crop. Farmers could achieve higher yields with early overall applications of glyphosate than with conventional herbicides, but with the same low weediness as conventional herbicides; alternatively, they could achieve equivalent yields to conventional herbicides with band sprays of glyphosate followed by late overall applications, but with the additional environmental benefits (insect food and habitat) from conservation of the weed flora for longer. This sets the FSE work in context as studying an environmental worst-case option for GMHT crop production systems. In this paper, we suggest that the way forward in row crops might be to use the technology to maximize environmental benefit and sustainability in a way that does not conflict with agronomic and financial benefits.

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